27 JULY 1979 EFFECTS OF NONIONIZING ELECTROMAGNETIC RADIATION (FOUO 3/79) 1 OF 1

JPRS L/8591

27 July 1979

USSR Report

BIOMEDICAL AND BEHAVIORAL SCIENCES

(FOUO 3/79)

EFFECTS OF NONIONIZING ELECTROMAGNETIC RADIATION

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INVESTIGATION OF THE EFFECTS OF A CONSTANT MAGNETIC FIELD ON ARTERIAL HEMODYNAMICS AT THE BASE OF THE BRAIN

Moscow BIOFIZIKA in Russian No 3, 1979 p 568

[Abstract of article by Ye. V. Zakharov and T. V. Peregudova, Moscow State University imeni M. V. Lomonosov; the complete article was filed on 5 Mar 79 with the All-Union Institute of Scientific and Technical Information, File No 805-79; submitted 15 Aug 76]

[Text] A new mathematical model describing the effect of a constant magnetic field on cerebral circulation is described. The model takes into consideration self-regulation of cerebral circulation; it singles out three functional groups of arteries that are capable of providing for this type of regulation independently of one another. Digital methods were used to analyze quantitative characteristics of the process (flow of blood to different segments of the arterial system of the base of the brain and blood pressure at arterial junctions).

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THE POSSIBLE ROLE OF WATER IN TRANSFER OF MILLIMETER RANGE RADIATION TO BIOLOGICAL OBJECTS

Moscow BIOFIZIKA in Russian No 3, 1979 pp 513-518

[Article* by S. A. Il'ina, G. F. Bakaushina, V. I. Gayduk, A. M. Khrapko and N. B. Zinov'yeva, Institute of Radio Engineering and Electronics, USSR Academy of Sciences, Moscow, submitted 8 Jul 77]

[Text] A study was made of the effect of radiation in the millimeter range (RMR) on human hemoglobin and blood erythrocytes, which is manifested by enhancement (or attenuation) of the heme-globin bond and decrease in osmotic resistance of erythrocyte membranes. The observed effect could not be attributed to mere warming of the irradiated suspension. A maximum effect on erythrocytes was obtained in a strongly diluted suspension, when the energy absorbed directly by erythrocytes is at a minimum. The obtained data warrant the assumption that the effects of RMR on biological objects are determined by interaction between the biological object and water in the radiation field, and not by the magnitude of directly absorbed energy.

Several studies [1-5] have demonstrated the existence of a nonthermal effect of RMR on biological objects on different levels of organization, including the hemoglobin (Hb) macromolecule. To explain these effects, it was assumed [4-7] that there is excitation of rotary oscillations of some dipole groups in the Hb active center, or else excitation of oscillations of protein as a whole was considered [8]. Thus far, however, there is no explanation for the fact that the magnitude of the effect is lower in the case of irradiation of powder than aqueous solutions of Hb. At the same time, with a low concentration of Hb in water ($c \approx 3-5\%$), as a result of considerable extinction of RMR in water, there is a relative decrease in RMR absorption by protein, the thickness of the irradiated layer being constant, with decrease in

*This paper was delivered at the All-Union Symposium on Instruments, Technology and Propagation of Millimeter and Submillimeter Waves in the Atmosphere, in "Tezisy dokladov i soobshcheniy" [Summaries of Papers and Reports], Institute of Radio Engineering and Electronics, USSR Academy of Sciences, 1976, p 316.

wavelength. Thus, the conception that the effects are determined by the field that permeates biological objects apparently requires more exact definition.

We conducted several experiments dealing with the effects of RMR on Hb and erythrocytes, in which: a) there was a substantial decrease in wavelength λ of radiation, as compared to other studies [2-5]; b) the concentration of erythrocytes in solutions was changed over a wide range.

Effects of RMR on aqueous solutions of Hb: It has been shown [2-5] that RMR (λ = 6-8 mm) has an effect on aqeuous solutions of Hb obtained from human erythrocytes; there was strengthening or weakening of the heme-globin bond. It was interesting to investigate the effects of shorter waves on Hb using the same method.

The Table lists data on the effects of RMR of $\approx$3 mW/cm^2. Hb was irradiated by a carcinotron ["backward-wave tube] (CT) on a unit of the quasioptical type. The Hb solution was decanted into teflon cuvettes, 1.5 mℓ in size, with a thickness of l = 1 mm. One of the cuvettes was in the RMR field and the other served as a control (Figure 1). The cuvettes were kept at a temperature of 37°C. Exposure time was 4 h. We determined the effect on the basis of dissociation of the bond between heme and globin [9].

Results of exposure to short mm range radiation	λ, мм	0,9	1,13	1,58	1,65	
	$\bar{\eta}\pm\sqrt{\sigma_\eta}$	1,1±0,03	0,83±0,08	1,17±0,11	1,02±0,09	
Data from [2-5] [2—5]	λ, мм	6,8	7,1	7,35	7,5	7,7
	$\bar{\eta}\pm\sqrt{\sigma_\eta}$	0,91±0,15	0,96±0,19	1,04±0,10	1,10±0,09	1,02±0,07

Note: $\bar{\eta}$ is the mean ratio of extent of HB dissociation in heme-globin bond in the experiment to extent of dissociation in the control. $\sqrt{\sigma_\eta}$ is the standard deviation.

To 2 mℓ 0.6% aqueous solution of Hb we added an aliquot of 1 N HCl, and the working mixture was incubated for 1 h at 30°. The sediment of dissociated heme was separated by centrifugation at 8000 r/min; the pH of the centrifugate was brought up to 6.8-6.85 with 0.1 and 1 N solutions of NaOH in the groove of an LPU-01 pH-meter; as a result of acid denaturation, the nondissociated Hb settled. After centrifugation and elution in phosphate buffer (pH 6.8), the obtained Hb sediment was dissolved in a minimum (2 mℓ) amount of 0.1 N NaOH solution. A series of dilutions was prepared from the obtained solution, to which we added the appropriate reagents to obtain a stained complex in the solutions (Lowry method). We determined the optical density of stained solutions using an SF-3A spectrometer at $\lambda = 656$ nm. Then we determined the true concentration of nondissociated Hb using a calibration curve plotted for an aqueous solution of oxy-Hb. We determined the extent of dissociation [splitting] X using the formula $X = (1-B/A)\cdot 100$, where A and B are the concentrations of initial and nondissociated Hb, respectively.

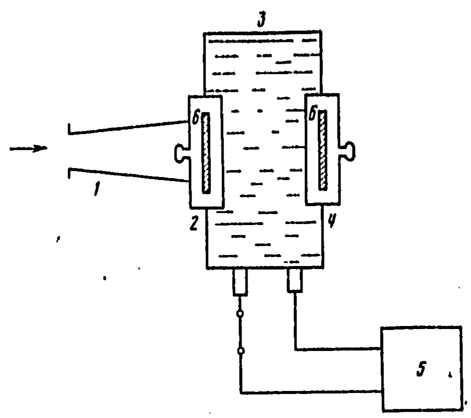

Figure 1.
Diagram of irradiation of Hb solutions and erythrocyte suspensions

1) funnel
2) irradiated cuvette (experiment)
4) nonirradiated cuvette (control)
3, 5) thermostats with water
6) Hb solution or erythrocyte suspension

The effect of RMR at $\lambda = 0.9 \div 1.65$ mm was found to approximately the same as at $\lambda = 6$-8 mm (see Table). At the same time, with $\lambda \simeq 1$, RMR penetrates only into the most superficial layer of the solution (extinction 40-50 dB/mm). Also of interest is the change in direction of the effect (see Table) at one of the wavelengths: at $\lambda = 1.13$ mm exposure leads to decreased stability of the heme-globin bond, whereas at other wavelengths there is an increase in stability of this bond.

Effects of RMR on osmotic stability of erythrocyte membranes: It is expedient to turn from examination of the effects on hemoglobin to the effect of RMR on Hb content of erythrocytes (E). At first, we studied the effect on a suspension of E of the same concentration.

We prepared a suspension of human E after removal of plasma, in approximately a 60-fold dilution (K = 60), as compared to the concentration of E in whole blood. The suspension was placed in the above-described curvettes. Exposure time constituted 3-3.5 h at 36-36.5°. Irradiated and control specimens were analyzed with an SF-4A spectrophotometer ($\lambda = 410$ nm). We measured the optical density of solution of Hb that migrated from erythrocytes into the surrounding medium (hypotonic solutions) [10, 11]. The degree of erythrocyte hemolysis was determined as a percentage of complete hemolysis, which was obtained by adding 0.1 mℓ suspension to 4.9 mℓ distilled water. The value of μ as a function of concentration of saline solutions *c* at three wavelengths (Figure 2, a–c) shows that RMR has an appreciable effect* on osmotic stability of E, in the direction of decrease.

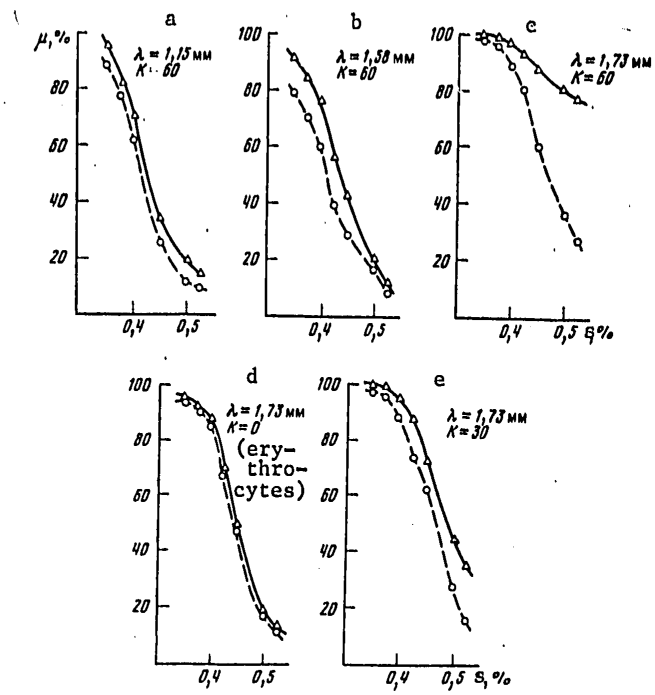

Figure 2. Function μ(*c*) for irradiated erythrocytes; uninterrupted line--experiment, dash line--control

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*Statistical processing [11] of the experimental results is indicative of reliability of the effect (E) of exposure. Indeed, the top and bottom ranges of the reliability intervals constitute $\overline{M} = E \pm m\rho_{1-p/2}$ and $\overline{m} = S/\sqrt{n}$ where S^2 is the unbiased estimate of variance for n experiments,

$S = \sqrt{\sum^{n}(\overline{E}-E_i)^2/(n-1)}$ and $\rho_{1-p/2}$ is the quantile of Student's distribution for the chosen level of significance $p = 0.05$; $\overline{E} = \mu_{exper} - \mu_{control}$. The difference in value of μ indicates that $\overline{M}>0$, i.e., the difference obtained as a result of irradiation in value of μ does not change in sign for all of the concentrations of erythrocyte suspension.

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In order to determine the influence of heating the irradiated specimens, we determined function $\mu(t)$ when the temperature in one of the cuvettes was raised by 0.5, 0.1 and 1.5° (Figure 3). A differential copper-constant thermocouple (d = 0.2 mm), graduated with a type M95 millivoltmeter on a scale of 0.5 mV was used for indication of heating. The effect of RMR is approximately equivalent to 1° heat. At the same time, according to the readings, heating did not exceed 0.1°, which is also confirmed by the theoretical estimates of heat. Consequently, the effect of RMR on permeability of E cannot be attributed to heating of the suspension.

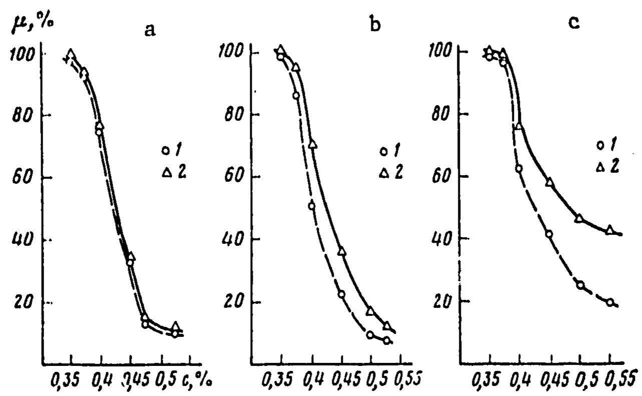

Figure 3. Function $\mu(c)$ for nonirradiated erythrocytes at different temperatures

1) t = 36.5°; a: 2) t = 37°, Δt = 0.5°
2) t = 37.5°, Δt = 1°; c: 2) t = 38°, Δt = 1.5°C

The heat estimates were made on the basis of theory and with the use of the programs in [12]; the numerical coefficients were taken from [13, 14]. Since there was heat emission in a thin layer (of the order of a skin layer), the thickness of which is small, as compared to the length of the cuvette, we solved the heat transfer equation of Laplace, with consideration of free convection (the source of radiation was on the surface) in order to estimate the maximum possible heating. With a layer of the order of 1 mm in thickness in the millimeter range of wavelengths, maximum heating constituted 0.1°, with a radiation flux density of $I \approx 1$ mW/cm^2 and 1° with $I \approx 10$ mW/cm^2, while the mean heating of the layer was several times less. A maximum temperature is observed near the surface of the cuvette oriented toward the radiation source. The time of establishment of a stationary state under the same conditions is 1÷10 s, which is much less than irradiation time. These estimates confirm the results of our measurements.

We irradiated E suspensions of different concentrations (Figure 2, c-e) at the wavelength of maximum effect (λ = 1.73 mm). No effect of RMR was demonstrated on eluated E (Figure 2d). With 15-fold dilution, the degree of hemolysis of irradiated E differs little from the value of μ for control suspensions. With K = 30, there is an appreciable decrease in stability of E membranes (Figure 2e). Finally, at K = 60 (Figure 2c), there is significant hemolysis of cells and, in a number of tests, total hemolysis of E.

Thus, our study of the effects of RMR on stability of the heme-globin bond in Hb in aqueous solutions and osmotic stability of E membranes containing Hb indicates that the magnitude of the effects are retained when solutions of Hb are used instead of dry preparations thereof, with reduction of wavelength, as well as an appreciable correlation between the level of nonthermal effect of RMR on E and concentration of the latter in the suspension. A maximum effect was obtained with strongly diluted specimens, when the overall energy directly absorbed by E is at a minimum. The obtained data warrant the assumption that the effect of RMR is not determined by the magnitude of absorbed energy, but by its interaction with the aqueous medium in the radiation field (for example, through the hydrate membrane of macromolecules and biological structures). For the time being, on the basis of the factual data, we can only assume that the effect of RMR on aqueous solutions of Hb and E suspensions is not related, to some extent, to the optical thickness of the irradiated specimen, i.e., it is determined by processes at the surface of the solution.

The described distinctions of RMR effects can be attributed, for example, to excitation in water of oscillations of the quasicrystalline (like ice) structure of water. We know of effects (used to measure force of RMR) of the mechanical action of coherent radiation on macroscopic objects with time-averaged force of radiation pressure. Perhaps, when biological objects are exposed to RMR transfer through water of an effect that is variable in time, i.e., that depends on the signal frequency, plays a role. The possibility of such an effect is attributable expressly to absorption of wave energy by the surface layer ($l_{skin} \ll l$). The variable force of radiation pressure $p(t)$ is determined only by its intensity I (I can reach several dynes/cm^2, which is higher by a factor of 10^4-10^5 than from noncoherent sources). It is rather important that the macromolecules themselves and other structures present resonance oscillations expressly in the millimeter and submillimeter ranges of waves [15]. Excitation of protein oscillations as a "solid body" in the case of macromolecules can apparently lead to "warming" of the active center, which is related to changes in biological structures that are manifested in experimental observation of the effects.

Thus, the possible scheme of effects of RMR on biological objects may be as follows:

Electromagnetic wave (RMR) falling on cuvette with solution	→	Superficial layer of water (skin layer, l_{skin}) in which RMR is absorbed	→	Excitation of oscillations in water and transfer thereof to substrate	→	Excitation in substrate of free oscillations and changes they induce

Of course, we cannot rule out the possibility that there is diffusion exchange of molecules or structures at the air-fluid interface. Since biological molecules or structures in the surface layer, which are slowly diffused from this layer are exposed to radiation, in this case prolonged exposure is required. The choice between a diffusion mechanism of exposure and mechanism of exposure "through water" should be based on additional data.

The demonstrated role of water in the effects of RMR can also explain why the data cannot be reproduced, to some extent, in dry specimens [2], on which the effect can indeed be at a maximum only when the powder-form specimens are moistened, since we know [16] that the properties of these preparations are critically related to the degree of hydration (vapor pressure above the samples). Evidently, the choice of optimum concentrations of solutions and suspensions is desirable in many cases, in particular in [17], where a resonance decrease was demonstrated in Hb yield from the irradiated mass after it was thawed.

The authors wish to express their appreciation to M. P. Petrova for her useful discussion of our study.

BIBLIOGRAPHY

1. Devyatkov, N. D. ELEKTRONNAYA TEKHNIKA [Electronics], series I: "SHF Electronics," No 4, 130, 1970.

2. Andreyeva, A. P.; Dmitriyeva, M. G.; Il'ina, S. A.; Koreneva, L. G.; Kud-yashova, V. A.; Merzlov, V. P.; Serbinova, T. A.; and Faleyev, A. S. Ibid, No 11, 12, 1971.

3. Kudryashova, V. A.; Il'ina, S. A.; Faleyev, A. S.; Gayduk, V. I.; and Dementiyenko, V. V. "Investigation of Resonance Effect of Millimeter Waves on Hemoglobin," preprint No 115, Institute of Radio Engineering and Electronics, USSR Academy of Sciences, Moscow, 1972; "Sb. materialov 4-go Vsesoyuznogo simpoziuma 'Gigiyena truda i biologicheskoye deystviye elektromagnitnykh voln radiochastot'" [Proceedings of 4th All-Union Symposium on "Industrial Hygiene and Biological Effects of Radio Frequency Electromagnetic Waves"], Izd-vo Institute of Industrial Hygiene and Occupational Diseases, USSR Academy of Medical Sciences, Moscow, pp 64, 65 and 112, 1972.

4. Gayduk, V. I.; Khurgin, Yu. I.; and Kudryashova, V. A. USPEKHI FIZ. NAUK [Advances in Physical Sciences], 110, 446, 1973; Ibid, "Scientific Session of the Department of General Physics and Astronomy (17-18 Jan 1973)," pp 452-469.

5. Devyatkov, N. D.; Gayduk, V. I.; and Kudryashova, V. A. in "Issledovaniya v oblasti radiotekhniki i elektroniki, 1954-1974 gg." [Research in 1954-1974 in the Field of Radio Engineering and Electronics], Moscow, IRE [Institute of Radio Engineering and Electronics], USSR AS [Academy of Sciences], I, 1974.

6. Koreneva, L. G., and Gayduk, V. I. DOKL. AN SSSR [Reports of the USSR Academy of Sciences], 193, 465, 1970.

7. Polukhin, A. T.; Il'ina, S. A.; and Kudryashova, V. A. "Estimation of the Effects of Weak High-Frequency Electromagnetic Radiation on Some Semifree Bonds in the Hemoglobin Molecule," Preprint No 120, IRE, USSR AS, Moscow, 1972.

8. Komov, V. P.; Shmelev, Yu. I.; and Chernavskiy, D. S. KRATKIYE SOOBSHCHEN. FIZ. [Brief Reports on Physics] (Physics Institute, USSR AS), No 9, 38, 1972.

9. Komov, V. P. "Isolation of Hemoproteins and Investigation of Some of Their Physicochemical Properties After Exposure to X-Rays," candidatorial dissertation, Leningrad, 1965.

10. Vasil'yev, P. S., and Petrova, M. P. in "Voprosy biofiziki, biokhimii i patologii eritrotsitov" [Problems of Biophysics, Biochemistry and Pathology of Erythrocytes], Krasnoyarsk, I, pp 20 and 302, 1960.

11. Il'ina, S. A., and Bakaushina, G. F. "Effects of Short Millimeter Radiation on Osmotic Stability of Erythrocyte Membranes," Preprint No 22 (202), IRE, USSR AS, Moscow, 1975.

12. Beluga, I. Sh., and Khaplanova, Z. I. ELEKTRONNAYA TEKHNIKA, Series I: "SHF Electronics," No 1, p 10, 1974.

13. Greber, G., and Erk, S. "Main Teaching on Heat Transfer," ONTI, Moscow--Leningrad, 1936.

14. Kutateladze, S. S., and Borishanskiy, V. M. "Handbook on Heat Transfer," Gosenergoizdat, Moscow--Leningrad, 1959.

15. Chernavskiy, D. S.; Khurgin, Yu. I.; and Shnol', S. E. MOL. BIOL. [Molecular Biology], 1, 419, 1967.

16. Khurgin, Yu. I. ZHURNAL VKhO IM. D. I. MENDELEYEVA [Journal of the All-Union Chemical Society imeni Mendeleyev], No 6, 684, 1976.

17. Zalyuboskaya, N. P.; Gordiyenko, O. I.; and Kiselev, R. I. PROBLEMY GEMATOLOGII I PERELIVANIYA KROVI [Problems of Hematology and Blood Transfusion], No 4, 31, 1975.

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EFFECT OF GENERAL ANESTHETICS ON MICE FOLLOWING MICROWAVE IRRADIATION

Moscow BYULLETEN' EKSPERIMENTAL'NOY BIOLOGII I MEDITSINY in Russian No 5, 1979 pp 425-427

[Article by V. M. Koldayev, Vladivostok Medical Institute]

[Text] There is a possibility that persons previously irradiated by microwaves may be subjected to general anesthesia in clinical practice, in surgery for example; however, the effect of such anesthetics on the irradiated organism is unknown.

This paper deals with the effect of some general anesthetics on survival of mice irradiated by microwaves, as well as with the time of onset and duration of general anesthesia following administration of the preparations different times after irradiation.

Research Methods

In experimental series I white mice (474) of both sexes (about 75 percent males) weighing 22-26 gm were irradiated by microwaves until a terminal condition was reached, as described earlier (1). Immediately after irradiation the mice were given a one-time dose of sodium thiopental--60 mg/kg, sodium oxybutyrate--10,50, and 500 mg/kg, hexenal--60 mg/kg, or urethane--800 mg/kg, intraperitoneally as aqueous solutions at a volume of 1 ml per 100 gm body weight; inhalational anesthetics were administered with air inhaled in a special 1 liter chamber containing a mixture of the anesthetic and atmospheric air at the follwoing ratios: fluorothane--1:10,000, chloroform--1:5,000, ethyl ether--1:1,000. The mice were kept in this chamber for 60 or 90 sec. We recorded mouse survival for 3 weeks after irradiation.

In experimental series II the mice were given one-time doses of ether, fluorothane, hexenal, or sodium thiopental different times following microwave irradiation--immediately after irradiation, and after 2, 4, and 10 days. We determined the time of onset and duration of collapse.

Irradiated mice that did not receive anesthetics served as the control in the survival experiments, while unirradiated mice receiving the anesthetics served as the control in experiments performed to determine the time of onset and duration of anesthesia. Eight to twelve mice were used in each experiment. The data were subjected to statistical treatment by the method of variation in qualitative signs (2).

Research Results

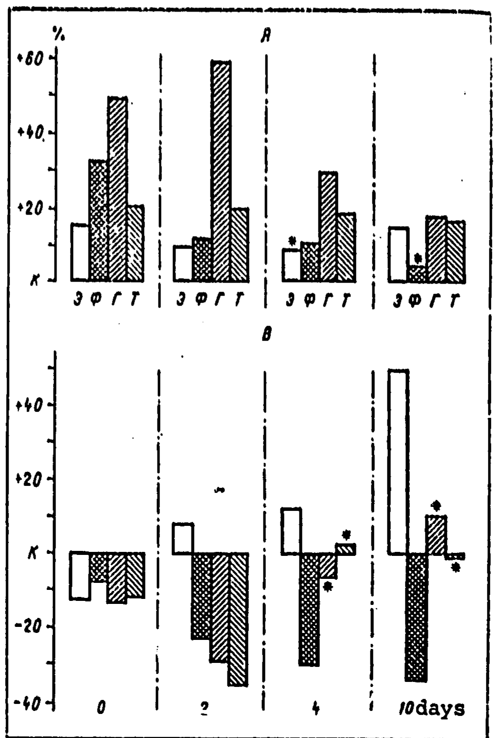
Survival of mice irradiated by microwaves decreased by 1.6-1.45 times in comparison with control ($P<0.05$) following administration of chloroform, fluorothane, sodium thiopental, and sodium oxybutyrate at a dose of 500 mg/kg, while in response to ethyl ether, hexenal, urethane, and sodium oxybutyrate at doses of 10 and 50 mg/kg the decrease in survival was insignificant ($P<0.05$; see Table).

Survival of Mice Receiving One-Time Dose of General Anesthetic Immediately After Microwave Irradiation ($M \pm m$)

Experimental Conditions	Survival, %	P
Control (67)	48 ± 6	—
Fluorothane, 90 sec (8)	30 ± 14	0.042
Chloroform, 90 sec (10)	30 ± 14	0.042
Sodium thiopental (12)	33 ± 14	0.036
Sodium oxybutyrate, mg/kg:		
10 (16)	43 ± 12	0.485
50 (8)	37 ± 17	0.310
500 (6)	33 ± 19	0.048
Ethyl ether, 60 sec (10)	40 ± 16	0.421
Hexenal (10)	40 ± 16	0.274
Urethane (12)	41 ± 14	0.319

Note: Number of mice is shown in parentheses.

The time of onset of collapse decreased by 8-14 percent in response to ether, fluorothane, hexenal, and sodium thiopental given immediately after irradiation; when administered later, ether caused a gradual 48 percent increase of this time in comparison with control, while fluorothane, administered 10 days after irradiation, caused the time to decrease by 34 percent. When hexenal and sodium thiopental were administered 2 days after irradiation the time of onset of collapse decreased by 28-36 percent, while when administered after 4 days the time of onset was the same as in control; the duration of collapse increased by 15-48 percent after these preparations were administered



Growth (Percent in Relation to Control) in Duration (*A*) and Time of Onset (*B*) of Collapse of Irradiated Mice Given Ether (Э), Fluorothane (Φ), Hexenal (Γ), and Sodium Thiopental (T) Different Times Following Irradiation: Abscissa--time after irradiation (days); ordinate--percent of control (*K*). Asterisk indicates $P>0.05$ in comparison with control

immediately after irradiation, while when they were administered on subsequent days this duration gradually decreased (see Figure).

Thus the experiments show that following microwave irradiation, the sensitivity of the body to general anesthetics apparently changes, which must be accounted for in anesthesiological practice when it becomes necessary to administer anesthetics to persons who had been subjected microwaves immediately or some time before anesthesia.

BIBLIOGRAPHY

1. Koldayev, V. M., BYULL. EKSPER. BIOL., No 3, 1976, p 285.

2. Rokitskiy, P. F., "Biologicheskaya statistika" (Biological Statistics), Minsk, 1973.

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THE BIOLOGICAL INFLUENCE OF HYPOGEOMAGNETIC ENVIRONMENTS

Moscow IZVESTIYA AKADEMII NAUK SSSR-SERIYA BIOLOGICHESKAYA in Russian, No 3, 1979, pp 342-353

[Article by V. I. Kopanev, G. D. Yefimenko and A. V. Shakula, S. M. Kirov Military Medical Academy, Leningrad, submitted 26 Apr 78]

This problem has been shown to be an urgent one in the epoch of scientific and technical revolution when the activities of many operators occurs in objects shielded from the magnetic field of the earth (GMF) 100-fold or more. For the purpose of studying the influence of the geomagnetic environment on warm-blooded animals a series of experiments was conducted on rabbits which had spent their entire embryogenesis and grown to the age of one month in a shielded chamber where the GMF was reduced to one-onehundredth of its normal level.

The results obtained confirm the necessity of the influence of the GMF for normal development of the living organism. In animals which had grown up in a reduced GMF, dystrophic disturbances were noted in the liver, cardiac muscle, and the gastro-intestinal tract. The plastic and energy metabolisms were disturbed, and acute suppression of the activity of the investigated enzyme systems, with the exception of the glycolytic system was demonstrated. Changes in the motor activity of the experimental animals and insufficiency of the neuromuscular apparatus were demonstrated. Mortality among the experimental animals was statistically higher than among the controls. In this article the authors raise the question of the necessity of further complex investigations of the demonstrated effects of the hypogeomagnetic environment.

[Text] The diversity of living organisms on Earth arose, evolved and exists in indissoluble unity with different factors of the external environment. Many of them are of an electromagnetic nature; therefore it is, in principle, possible that any of the ranges of the electromagnetic spectrum of the biosphere played a role in the evolution of nature (Parin, 1968). In A. S. Presman's opinion, (1974), in the process of evolution nature used natural electromagnetic fields as a source of information facilitating the continuous adaptation of living organisms to the conditions of the external environment.

One of the components of the electromagnetic spectrum of the biosphere is the geomagnetic field (GMF), the important role of which in the life activities of man and of different biological subjects has been quite fully treated in A. P. Dubrov's monograph (1974). In this age of scientific and technical revolution, a person working as an operator in special buildings isolated from the external environment may be in a hypogeomagnetic condition. According to Wever's data (1967), concrete buildings have shielding properties which reduce the intensity of the GMF to one-onehundredth or less of its normal level. Moreover, as a result of space research it has been determined that the magnetic fields of the Moon, Mars, Venus and interplanetary space are significantly less than the magnitude of the GMF (Gringauz et al, 1960; Pochtaryëv et al, 1966).

Thus, investigation of the biological influence of hypogeomagnetic environments is urgent not only in its evolutionary aspect, but also for labor physiology, space biology and medicine and in terms of philosophy.

At the present time, the literature includes approximately 60 studies devoted to the influence of hypogeomagnetic environments on biological subjects. The majority of them were carried out on microorganisms and plants, a fact which makes extrapolation of the results obtained to higher animals and man extremely difficult. The elementary works of American authors (Halpern, Van Dyke, 1966; Busby, 1968; Conley, 1969; Beisher, Grisset, 1971) on investigation of the biological effect of hypogeomagnetic environments on the developing organisms of mammals are contradictory.

Taking into account the theoretic and practical significance of the question of the influence on mammals of hypogeomagnetic environments, the fact that the problem has been little studied and the contradictory nature of the data in the literature, we conducted a complex investigation, the objects of which were the following: first, to study the behavioral and certain physiological reactions of young rabbits which had undergone embryonic and postnatal development in a hypogeomagnetic environment; second, to demonstrate morphological changes in the vital internal organs; third, to investigate the activity of key enzymes in the brain, liver and stomach.

Methodology of the Investigation

Studies were conducted on young rabbits one month old obtained from female rabbits which were taken in pairs from one nest and covered by the same male. The experimental females were placed in a chamber shielded from the GMF, where pregnancy, birth and the development of the young rabbits to the age of one month occurred.

In order to shield the animals from the GMF and electromagnetic fields of industrial origin a multi-layer cylindrical screen consisting of four casings was developed and manufactured in the VNII [All-Union Scientific Research Institute] of Metrology imeni D. I. Mendeleyev. The three internal casings were made of Permalloy and were designed to screen from continuous low-frequency alternating (up to several hertz) magnetic fields. The fourth (external) casing was made of copper sheeting and was designed to shield from electromagnetic fields of industrial origin. The operating capacity of the chamber is 0.15 m^3. The magnetic field was measured with a nanoteslometer with an iron probe. The GMF was reduced to one-sixhundredth its normal level.

During the entire pregnancy and the development of the young rabbits to the age of one month the control rabbits remained in conditions of the usual GMF at the latitude of Leningrad. Cage dimensions, temperature, humidity, the light regime, feeding and other conditions were identical in the groups of experimental and control animals. The experiments were conducted during the period March to December 1976.

The course and duration of pregnancy in the female rabbits was observed taking into consideration the dates of nest-building and the birth of the young rabbits, their number at birth, mortality and behavioral reactions during the month after birth. We studied the growth dynamics of the young rabbits, weighing them on the 5th, 12th and 26th days after birth.

The motor activity of the young rabbits was investigated using a device of our creation. The neuromotor activity of the young rabbits was investigated using a M40/A four-channel myographic measuring system. One month after birth the animals were decapitated. Pieces of the internal organs were fixed in a 10-percent formalin solution. Sections 5 μm thick were stained with hematoxylin-eosin and in order to demonstrate lipds, with sudan III. Histochemical investigation of the mucosa of the stomach and the duodenum included study of: mucin production (Schick reaction and alcyanine blue); the functional activity of the secretory elements of the glands and the integumentary epithelium using Fel'gen's method of DNA staining; RNA using Brash's method; the protein-lipid complex using Berenbaum's method (Pirs, 1962); mast cells by staining the sections with basic brown according to M. G. Shubich et al (1973); and HCl using toluidine blue with subsequent acid hydrolysis; identification of gastrin-producing (G-), histamine-producing (EcL-) and serotonin-producing (Ec-) cells using Grimelius, Sev'er-Munger and Masson's method (Shubich et al, 1973) of silver staining with subsequent morphometry of the cells in a 1 mm^2 area of the section. The thickness of the mucous membrane was measured, the secretory elements of the fundal glands (main, accessory and lining cells) were counted, and the "mean fundal glands" determined using V. A. Samsonov's (1975) formulae.

The activity of the following key enzymes was demonstrated in the brain by histochemical methods: the citric acid cycle--succinate dehydrogenase (SDH) by E. Pirs' method (1962); the pentose cycle--glucose-6-phosphate dehydrogranse (G-6-PDH) by E. Pirs' method (1962); tissue respiration--NAD.N_2-dehydrogenase using M. Berston's method (1965); glycolysis--hexokinase (HK) (Siegel, Pette, 1969).

The activity of these enzymes was evaluated according to V. I. Kolodin and O. K. Kuznetsov's method (1975).

Determination of ATPase activity was performed by the method described (Wittam, Ager, 1964). Lactate dehydrogenase activity (LDH) was determined by G. A. Kochetov's (1971) spectrophotometric method.

Determination of glucose-6-phosphate dehydrogenase (G-6-PDH) was conducted according to the well known method (Kornberg, Horecker, 1955). Glutamate dehydrogenase (GDH) activity was determined by the method of V. M. Yakovlevoy et al (1964).

Altogether 965 investigations were conducted (table 1). The results obtained were analyzed using Student's t-test on a Wang electronic computer and the Wilcoxson-Manna-Whitney U-test (Gubler, Genkin, 1973).

Results and Evaluation

General Description of the Development and Certain Physiological Reactions of Young Rabbits

The course of pregnancy and the behavior of the experimental female rabbits did not significantly differ from those of the controls. The number of young rabbits was approximately equal at birth, but subsequent mortality in the group of experimental animals was 30 percent higher on the average compared to the controls ($P < 0.05$). The mean weight of the experimental animals was also significantly higher during the entire period of observation (table 2).

Moreover, the distribution of weight among the experimental and control groups was significantly different. The coefficient of weight variation of the experimental rabbits was 17 percent versus 8 in the control group, the indicated relationship persisting throughout the entire experiment.

Flaccidity and low mobility were characteristic of the experimental animals. When they were isolated from the nest, however, motor activity increased by 59 percent ($P < 0.05$, n=24) compared to the control animals. In addition, the experimental young rabbits constantly tried to leave an enclosed space and return to their own nest, something that was not noted in the control animals.

The differences determined took place also in the qualitative description of movement. The young rabbits of the control group were constantly in motion, their movement practically not differing in amplitude. In the young rabbits of the experimental group, however, short periods of intensive movement alternated with prolonged periods of almost complete immobility.

Investigation of the neuromotor activity (table 3) showed that in young rabbits of the experimental group, the threshold of sensitivity of the sciatic nerve was decreased by 36 percent compared with the control group ($P < 0.05$) and the latent period of muscle response, to the contrary, was increased by 40 percent ($P < 0.001$). The latter circumstance may be a result not only of the decrease in the frequency of conduction of stimulation along the nerve fiber, but also of the increase in the lag in impulse conduction in the neuro-muscle synapse.

Along with the change in the threshold of sensitivity of the peripheral nerve and the latent period the amplitude of muscle contractions increased 1.51-fold, a result which apparently is caused by the involvement of a greater number of muscle fibers in the response reaction in young rabbits developing in a hypogeomagnetic environment.

Results of Morphological Investigations

In the morphological studies a tendency to adiposis in the region of the kidneys, heart, and abdominal cavity was distinctly demonstrated macroscopically in the experimental animals. The dimensions of the internal organs in the experimental rabbits exceeded the dimensions of the controls. The liver of the experimental animals had a pronounced yellow hue; in several animals there were cirrhosis-like changes expressed in the manifestation of tuberosity and induration of heptic tissue.

Microscopic investigation of the liver of the experimental animals (hematoxlin-eosin stain) showed that the lobular structure is preserved in the liver, and the peripheral parts of the lobes stain brightly oxyphilic consisting of cells with dense acidic cytoplasm. Closer to the central vein numerous oxyphilic granules are demonstrated in the cytoplasm of the hepatocytes (the cytoplasm itself becomes less oxyphilic). In the central region of the lobule the cells are strongly vacuolized the cytoplasm of the vacuoles; is forced back to the cellular membrane and connected to the nucleus by fine, bright streaks. In the hepatic lobes, the capillaries are in spasm and indistinguishable in places. The Kupffer cells are reactive. Their nuclei look large and disintegrating,and the cytoplasm looks extremely dense and thickened, possibly edematic. The endothelium of the veins, particularly their subendothelial layer, is changed reactively.

There is reason to suppose that not only the RES [reticulo-endothelial system] cells but also the hepatic tissue is reactive. This is indicated by the numerous mitoses in the hepatocytes and the presence of flattened, degenerating cells with lysed nuclei.

In some of the animals a somewhat different picture was observed: most of the cells were vacuolized; the cytoplasm was forced back by the vacuoles toward the periphery and was mildly oxyphilic, hepatic gullies were encountered, especially at the peripheral lobules where lysis of all the hepatic cells and expecially their nuclei is distinctly apparent (only shadows of hepatocytes are encountered). In these cases, the predominance of the dystrophic changes of hepatocytes is evident. There are few Kupffer cells; they are reactive, and destructive changes predominate. The interlobular bile ducts have thickened external connective tissue membrane. Connective tissue grows out from the ducts into the hepatic lobules, apparently on sites of the dead gullies. Bands of young hepatocytes emerge from the bile ducts. These bands, however, are short and do not reach even the external third of the lobe. In these conditions it is possible to speak of the prevalence of the dystrophic processes in the liver (fig 1). Accordingly, we can speak either of two possible paths of reaction of the liver in a hypogeomagnetic environment, or, what is more likely, about two stages of one process in which at first general stimulation not only of the liver cells but also of the reticulo-endothelial system cells is observed, and during the second stage the dystrophic changes develop which are characterized by vacuolization and lysis of the hepatocytes, destructive changes in the Kupffer cells, and proliferation of connective tissue. The dystrophic nature of the reaction is confirmed by staining with sudan III. The majority of the liver cells contain lumps of fat; cells completely filled with fat and containing no nuclei are encountered (fig 2).

When cardiac preparations are stained with hematoxilin-eosin, the phenomenon of dystrophy is observed in the experimental animal; it was manifested in the decrease in the quantity of myofibrillae, pyknosis and the destruction of nuclei. The dystrophic manifestations are more pronounced toward the periphery. Sections with acute polymorphism are encountered, where along with dead and dystrophic cells there are young, short cells, the growth of which is inconsistent with the radial distribution of the muscle fibers.

There are few endothelial cells; their nuclei, which are enlarged, acquire an ovoid form not normally characteristic of them. The nuclei which maintained their usual thickness increased 2-2.5-fold in length. The cells of the subendothelial layer are in a condition of pronounced reactivity. Consequently, in the cardiac muscle acute manifestations of dystrophy increasing toward the periphery and polymorphism of the structure of cardiac muscle are demonstrated. The epithelium of the capillaries, especially their subendothelial layer, is changed reactively.

In the spleen there are signs of intenstification of blood circulation and the number of lymphoid elements in the red pulp and Malpighian bodies is increased. A small degree of differentiation of the lymphod elements is noted; the majority of lymphocytes are large.

In the lungs the basic changes involve the respiratory tracts, especially the small passages. Single lymphoid follicles are encountered in the small bronchia, and active secretion of the mucous cells of the apocrine type is noted.

The results of histochemical and morphometrical investigation of the mucous membrane of the stomach and duodenum are presented in Table 4 which shows that in the experimental young rabbits a small decrease in nearly all indices took place in comparison with the controls. In particular, the thickness of the mucous membrane of the body of the stomach was smaller by 29.5 percent in the anterior region--29.5 percent smaller and in the duodenum--50 percent smaller. The decrease in the thickness of the mucous membrane was caused mainly by shortening of the villi.

Mast cells were distributed equally in the mucous membrane of the stomach and the duodenum, however, in the experimental rabbits their quantity in the gastric mucosa was decreased by 62.3 percent; by 25.2 percent in the external section and the pyloric canal; by 50 percent in the duodenum. Predominance of rounded cellular elements with unsaturated granules in the cytoplasm was also noted.

There were also significant differences in comparing the endocrine cells of the gastroduodenal mucosa. Thus, in comparison with the controls, there were significantly fewer G-cells in the experimental animals (in the mucosa) of the body of the stomach--87.2 percent, in the external sections 72 percent, and in the duodenum 41.2 percent fewer), and especially fewer EcL-cells which in the experimental rabbits were absent in the mucosa of the body of the stomach and the duodenum and were found only individually in the mucous membrane of the external sections. An insignificant decrease in Ec-cells in the mucosa of the body of the stomach was also discovered in the experimental rabbits compared with the controls, but a 1.5-fold increase was demonstrated in the external sections of the stomach, and an 11-fold increase, in the mucous membrane of the duodenum.

In this way, in rabbits developing in a hypogeomagnetic environment, pronounced changes take place in the mast cells and in the endocrine apparatus of the mucous membrane of the stomach and duodenum. Decrease in all sections of the gastroduodenal system of gastrin- and histamine-producing cells is characteristic; it takes place against the background of atrophy of the mucous membrane. The atrophic process apparently is the result of decrease in the regenerative capacity and disturbances in the process of differentiation of cellular elements. The latter finding graphically confirms the data of Table 5, which presents the results of morphometry of the fundal gland cells.

Table 5 shows that disturbance of the differentiation of cellular elements of the main gastric glands is observed in the course of embryogenesis and the development of the young rabbits during the month after birth in a hypogeomagnetic environment. This is manifested as a decrease in the highly differentiated main and lining cells and an increase in the less differentiated accessory cells. Such reorganization of the main gastric glands results in "mucoidization" of the latter and "mucusization" of the stomach.

Based on serotonin's properties which stimulate mucin-production, the process of the "mucoidization" of the mucous membrane of the stomach apparently also promotes an increase in Ec-cells in the lower section of the stomach and in the duodenum.

The decrease in the number of lining cells may also be caused by the reduction of the gastrin mechanism for stimulating the main gastric glands. The weakening of the stimulating effect of gastrin and histamine is borne out by a decrease in RNA and DNA in the main cells and of the protein-lipid complex in the lining cells; the weakening of their trophic effect is confirmed by a decrease in the number of these cells.

Results of Histochemical Investigation of Key Enzymes in the Brain

The cortex and regions of the associative and comissural pathways of the brain of the rabbits were subjected to histochemical study. In the core of the brain the perikaryon of neurons--predominantly of three-four layers--and astroglial cells were analyzed; in the white substance, the oligodendroglial elements. The results of the investigation are presented in table 6.

In the bodies of the nerve cells of layers 3-4 of the brain of the control animals, very high G-6-PDH and NAD.N_2-DH activity, moderate SDH and weak HK activity were determined. When the reaction to G-6-PDH was analyzed, it was noted that the intensive formation of the granules of diformazan was characteristic not only of the neurons of cortex but also for the cells of the astroglia and the oligodendroglia. Increase in the deposition of diformazan was documented by the presence of very dense distribution of dark-blue granules in the cytoplasm of the cells and their processes. Moreover, the diformazan granules were localized in the fibers of the conducting pathways of the white matter of the brain. In some cases in sections of the brain, fragments of the choroidal plexus of the cerebral ventricles, epidermal, and subepidermal layers were detected in which G-6-PDH activity was also rather high. NAD.H_2-DH activity, as a rule, was high in the perikaryon of the cells of the cerbral cortex and moderate or pronounced in the astroglia and oligodendroglia. In the nerve fibers deposition of diformazan granules atypical in form and varying in hue was also noted.

On demonstration of SDH moderate deposition of diformazan granules was found not only in the body of the cells of the cerebral cortex belonging to the pyrimidal and ganglionic layers, but also in the cytoplasm of astrocytes of the cytoplasmic type. The granules were small and brownish-lilac in color. The reaction to SDH in the elements of the oligodendrolia was relatively low.

The level of HK activity in the cellular elements investigated was low with the exception of the endothelium of the capillaries where in three cases the strength of the key enzyme of glycolysis reached a pronounced degree of activity.

Thus, very high G-6-PDH activity and low GK activity were noted in the brains of the control animals. This result indicates the predominance of the phosphorylase pathway for replenishing the intra-cellular stock of glucose-6-phosphate and the significant contribution of the pentose pathway in utilization of the intermediates of carbohydrate metabolism. The results obtained coincide with G. S. Khachatryan's data (1967) where high G-6-PDH activity was detected in brain tissue. High $NAD.N_2$-DH and moderate SDH activity were also noted, indicating the predominance of the $NAD.N_2$-dependent pathway for transfer of restorative equivalents to the respiratory chain and the definite contribution of the SDH system to this process.

In the perikaryon of the nerve cells of layers 3-4 of the cerebral cortex of the experimental animals, the reactions studied were distributed in the following descending order by intensity of formation of diformazan granules: $NAD.N_2$-DH, G-6-PDH, SDG and HK.

$NAD.N_2$-DH activity was characterized by deposition of dark-blue small uniform diformazan granules which were evenly distributed in the bodies of the cerebral cortex nerve fibers. When G-6-PDH was demonstrated, the deposition of diformazan was greater in the pyrimidal cells and less pronounced in the molecular, external granulated and polymorphous layers of the cortex. SDH activity was weak, sometimes moderate, in all layers of the cortex, the diformazan granules were small, uniform, brownish-lilac. GH activity was detected in the form of traces. In the macroglia intensive formation of diformazan granules was noted on investigation of $NAD.N_2$-DH. The reaction to G-6-PDH showed moderate activity. Astrocyte activity was very low. The oligodrendrial elements distributed along the nerve fibers of the white substance of the brain had pronounced G-6-PDH and $NAD.N_2$-DH activity and uniform small dark-blue diformazan granules were distributed in the cytolasm of the oligodendrocytes. The granules were practically indistinguishable in the dendrites of the neurons. The subsequent reaction was characteristic of the cytoplasm of the oligodendriocytes when SDH and HK were demonstrated.

In this way, in the brain of experimental animals a significant decrease in G-6-PDH activity was noted, while HK activity remained on its former low level. This indicates the suppression of the pentose pathway of utilization of glucose-6-phosphate in hypogeomagnetic environments. This may, in turn, be caused either by accumulation of intermediates of the pentose pathway which inhibit G-6-PDH activity or by decrease in requirement for $NAD.N_2$-DH and ribose synthesis. A moderate decrease in $NAD.N_2$-DH and SDH activity was also demonstrated, which may indicate a total decrease in the metabolic activity of cellular elements of the cerebral cortex in experimental rabbits, inasmuch as the activity of these enzymes to a significant degree reflects the intensity of the occurence of the Krebb's cycle reaction--the central metabolic hub of the cell.

The Results of Biochemical Investigation[2]

The investigation conducted showed that total ATPase activity in the liver and the mucous membrane of the stomach decreased in the group of experimental animals. Expecially pronounced changes occurred in the gastric mucosa where ATPase activity decreased by 50 percent ($P < 0.01$, n=36), while in the liver it decreased by 28.6 percent ($P < 0.001$).

The data obtained also indicate a decrease in G-6-PDH and GDH activity. In the experimental group G-6-PDH activity decreased in hepatic tissue by 49.5 percent ($\rho < 0.01$); by 48.2 percent in the gastric mucosa ($\rho < 0.001$). The decrease in GDH activity was 30 percent in the mucous membrane of the stomach ($\rho < 0.01$) and 36.4 percent in the liver ($\rho < 0.01$). The decrease in GDH activity--one of the main enzymes of amino acid metabolism--is an indication of the disturbance of the plastic metabolism in the hypogeomagnetic environment.

Against the background of decreased activity of many enzymes, the increase in LDH--a glycolytic enzymes--is interesting. In the group of experimental animals, LDH activity increased by 22.3 percent in the liver ($\rho < 0.01$) and by 14 percent in the mucous membrane of the stomach ($\rho < 0.01$), a fact which indicates the intensification of anaerobic glycolysis.

The data obtained shed light on the results of the physiological and morphological studies. It can be conjectured that disturbance of the regulation of protein synthesis is at the root of many disturbances of plastic and energy metabolism, a hypothesis which is indicated by the decrease in DNA and RNA in the main cells of the gastric glands.

Investigations of the general motor activity of the rabbits showed a significant decrease in it in the group of experimental animals. As is well known, ATP is a necessary component of muscle contraction; its chemical energy is transformed into mechanical energy in the process of muscle contraction. The results obtained indicate an decrease in ATP production in the experimental animals which is apparently also caused by the decrease in general motor activity. ATP is also necessary for functioning of K^+/Na^+ -ATPase activity and reducing the intracellular level of potassium.

The complex investigation conducted on the biological effect of hypogeomagnetic environments on the developing organism of mammals has shown that the hypogeomagnetic environment is a biologically active factor. Proof of this are the facts we obtained in investigating the rabbits at different levels (from the organism to the molecular level) using physiological, morphological and biochemical methods.

First, in the groups of animals which developed under hypogeomagnetic conditions, increase in mortality, disturbance in the growth process, decrease in total motor activity and reduction of the threshold of excitability of the sciatic nerve were noted.

Second, in the experimental animals dystrophic changes in the liver and myocardium, pronounced disturbances of the mast cell and endocrine apparatuses in the mucous membrane of the stomach and duodenum and disturbances of cell differentiation in the myocardium and mucous membrane of the stomach were demonstrated.

Third, a decrease was detected in the activity of key enzymes of the Krebb's cycle, the pentose cycle and the plastic metabolism of the vital organs of

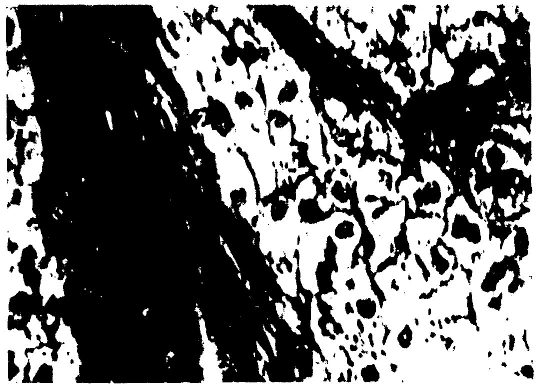

Figure 1. Outgrowth of intralobular connective tissue in the liver of experimental animals, lysis and pyknosis of the nuclei (hematoxylin-eosin stain, ocular 10, objective 12.5).

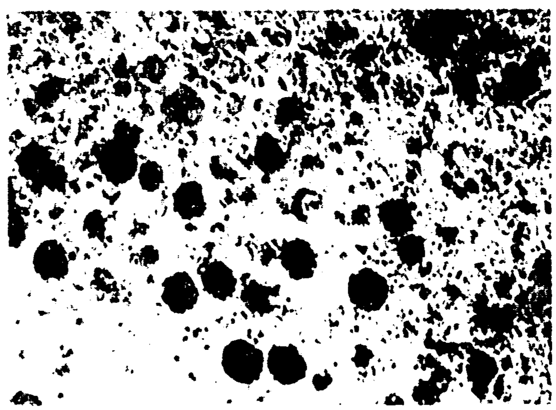

Figure 2. Fatty degeneration of hepatic tissue (sudan III stain, ocular 10, objective 12.5).

Table 1. Total Number of Studies

Type of Study	Number of Studies: control	Number of Studies: experimental
Physiological studies:		
Weight: 5th day	16	16
12th day	16	16
19th day	16	16
26th day	16	16
Motor activity	24	24
Neuromotor activity	48	48
Total	272	
Morphological studies:		
Liver	88	88
Heart	27	28
Spleen	5	6
Lungs	5	6
Stomach and duodenum	48	48
Total	349	
Histochemical studies of enzymes of the brain:	28	28
Total	56	
Biochemical studies:		
G-6-PDH	36	36
HDH	36	36
LDH	36	36
Total ATPase	36	36
Total	288	
Total number of studies:	965	

Table 2. Weight Dynamics of the Young Rabbits During the Investigation

Age, (days)	Mean Weight of Rabbits, g M±m at n=32		Age (days)	Mean Weight of Rabbits, g M±m at n=32	
	Control	Experimental		Control	Experimental
5	59,42±3,42	65,25±7,24	19	182,92±4,19	[illegible],18±9,42
	$P=0,05$			$P<0,001$	
12	124,35±3,14	151,93±8,43	26	294,35±7,43	348,31±24,18
	$P=0,004$			$P=0,039$	

Table 3. Results of Investigation of Neuromotor Activity of Young Rabbits

Indices	M±m at n=24		Indices	M±m at n=24	
	Control	Experimental		Control	Exptl.
Threshold of excitability, g	22±1	14±3	Duration of response reaction at 2 thresholds, msec	3,0±0,25	2,25±0,50
	$P<0,05$			$P<0,05$	
Latent period, msec	0,31±0,06	0,50±0,03	Amplitude, mV	0,56±0,10	1,16±0,20
	$P<0,001$			$P<0,05$	

Table 4. Morphometric and Histochemical Indices of the Mucous Membrane of the Stomach and Duodenum

Localization of mucous membrane	Thickness of mucous membrane, m		Mast cells/mm^2		Endocrine cells/mm^2: G-cells		EcL-cells		Ec-cells	
	Control	Exptl.	Control	Exptl.	Control	Exptl	Control	Exptl	Control	Exptl
Body of stomach	88 ± 4	62 ± 4	508 ± 115	243 ± 45	172 ± 52	22 ± 16	248 ± 62	0	174 ± 8	152 ± 51
	$p < 0.05$		$p < 0.01$		$p < 0.01$		$p < 0.01$		$p < 0.05$	
Lower Region of stomach and pyloric canal	73 ± 5	52 ± 3	313 ± 28	234 ± 58	265 ± 33	74 ± 11	369 ± 79	8 ± 3	179 ± 32	277 ± 37
	$p < 0.05$		$p < 0.05$		$p < 0.01$		$p < 0.05$		$p < 0.05$	
Duodenum	156 ± 10	78 ± 3	880 ± 125	444 ± 75	325 ± 84	191 ± 72	279 ± 91	0	48 ± 28	541 ± 50
	$p < 0.05$		$p < 0.01$		$p < 0.05$		$p < 0.05$		$p < 0.01$	

Table 5. Formula of Main (Fundal) Gastric Glands

Cells	Control	Exptl	Cells	Control	Exptl
Main	40±3	11±1	Accessory	27±4	108±8
	$P<0,01$			$P<0,01$	
Lining	160±12	101±6	Total	227±31	220±28
	$P<0,05$			$P>0,05$	

Table 6. Enzyme Activity in Different Types of Brain Cells

Type of Cells	SDH		G-6-PDH		NAD.N_2-DH		HK	
Investigated	Control	Exptl	Control	Exptl	Control	Expt	Control	Exptl
Body of Neuron	+ +	+	+ + + +	+ +	+ + + +	+ +	+	±
Astroglia	+ +	±	+ + +	+ +	+ + +	+ +	+	±
Oligodendroglia	+	±	+ + + +	+ + +	+ + +	+ +	+	±

animals which had spent their embryonic and postnatal development in a hypogeomagnetic environment. It is possible that under these conditions disturbance of protein synthesis occurs, and quantitative and negative qualitative changes take place in the enzymes as a result.

At the present time we do not have the facts which might explain how and at what level the hypogeomagnetic enviroment exerts its influence on the protein-synthesizing apparatus of the cell; however, the nature of the changes detected make it possible to hypothesize that the presence of the GMF is necessary not only for indirect hormonal but also for direct cellular regulation of the protein-synthesizing processes.

At present it is not clear at what stage of the animal's development the hypogeomagnetic field begins to exert its influence and what would be the result of animals' staying longer in hypogeomagnetic conditions. The experimental data obtained undoubtedly indicate the necessity of further study of the biological influence of hypogeomagnetic environments in order to demonstrate the mechanism of the influence of the given factor and to develop prophylactic remedies aimed at preventing deleterious shifts in the organism.

FOOTNOTES

1. N. V. Zgoda participated in conducting the experiments.

2. I. Sh. Galeyev participated in conducting the experiments.

BIBLIOGRAPHY

1. Berston, M. "Gistokhimiya fermentov [Histochemistry of Enzymes], Moscow, Mir, 1965.

2. Gringauz, K. I.; Bezrukhikh, V.V.; Ozerov, V.D. Rybchinskiy, R.E. DOKL AN SSSR, Vol 131, 1960.

3. Gubler, Ye. V.; Genkin, A.A. "Primeneniye neparametricheskikh kriteriyev statisticheski v mediko-biologicheskikh issledovaniyakh" [Statistical Use of Nonparametric Criteria in Medico-Biological Investigations], Leningrad, Meditsina, 1973.

4. Dubrov, A. P. "Geomagnitnoye polye i zhizn'" [The Geomagnetic Field and Life], Leningrad, Gidrometeoizdat, 1974.

5. Kolodin, V. I.; Kuznetsov, O. K. VOPR ONKOLOGII, vol 21, 1975.

6. Kochetov, G. A. "Prakticheskoye rukovodstvo po enzimologii" [Practical Guide to Enzymology], Moscow, Meditsina, 1971.

8. Parin, V. V. Foreword to: A. S. Presman, "Elektromagnitnyye polya i zhivaya priroda" [The Electromagnetic Field and Nature], Moscow, Nauka, 1968.

9. Pirs, E. "Gistokhimiya. Teoreticheskaya i prikladnaya" [Theoretical and Applied Histochemistry], Moscow, Izd-vo inostr lit, 1962.

10. Pochtarev, V. I. "Magnetizm Zemli i kosmicheskogo prostranstva" [The Magnetism of the Earth and Interplanetary Space], Moscow, Nauka, 1966.

11. Presman, A. S. "Elektromagnitnaya signalizatsiya v zhivoy prirode" [Electromagnetic Signalling in Nature], Moscow, Nauka, 1974.

12. Samsonov, V. A. "Yazvennaya Bolezn'. Novyye materialy k patomorfologii oslozhnennykh yeyё form" [Peptic Ulcer. New Materials on the Pathomorphology of its Complicated Forms], Petrozavodsk, Izd-vo Petrozavod in-ta, 1975.

13. Khachatryan, G. S. "Biokhimiya golovogo mozga pri normal'nykh fiziologicheskikh usloviyakh. Geksozomonofosphatnyy shunt v mozgu" [The Biochemistry of the Brain in Normal Physiological Conditions. The Hexosemonophosphate Shunt in the Brain], Yerevan, Yastan, 1967.

14. Shubich, M. G.; Lagynova, Zh. K.' Lutsenko, N. M. "Udebno-metodicheskoye posobiye po vnedreniyu dostizheniy gistokhimii v uchebnyy protsess na kafedrakh gistologii meditsinsikh institutov" [An Educational and Methodological Handbook on Application of the Achievements of Histochemistry in the Educational Process in the Histology Departments of Medical Institutes], Krasnodar, 1973.

15. Yakovleva, V. M.; Kretovich, V. M.; Gil'manov, M. K. BIOKHIMIA, Vol 29, 1964.

16. Beisher, D. E.; Grissett, J.D. "Aerospace Medical Association, Annual Scientific Meeting, Preprints", Houston, Texas, 1971.

17. Busby, D. E. SPACE LIFE SCI, Vol 1, No 1, 1968.

18. Conley, C. C. In: "Biological Effects of Magnetic Fields", NY, Plenum Press, 1969.

19. Kornberg, A.; Horecker, B. "Methods in Enzymology", Vol 1, NY, 1955.

20. Halpern, M. H.; Van Dyke, J. H. Abstract Scientific Program Aerospace Medical Association, 37th Annual Scientific Meeting, Las Vegas, 1966.

21. Sigel, P.; Pette, D. J HISTOCHEM CYTOCHEM, Vol 17, No 4, 1969.

22. Wever, R. J VERGLEICH PHYSIOL, Vol 56, No 2, 1967.

23. Wittam, R.; Ager, M. J BIOCHEM, Vol 93, No 2, 1964.

9380
CSO: 18/+0

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THE RESPONSE OF BEES TO ELECTRIC FIELDS

Moscow IZVESTIYA AKADEMII NAUK SSSR-SERIYA BIOLOGICHESKAYA in Russian No 3, 1979, pp 395-400.

[Article by Ye. K. Yes'kov and A. M. Sapozhnikov, NII [Scientific Research Institute] of Apiculture, Rybnoye, Ryazan'skaya Oblast', submitted 16 Jan 78]

The minimum intensity of an electric field (EF) in the frequency range of 100 to 1000 Hz which stimulates the first indices of excitation of bees is 5-14 V/cm. Under the influence of an EF of several dozen volts per centimeter strong excitation of a colony involving change in the microclimate of the hive is observed. Other things remaining equal, continuous electric oscillations have the greatest effectiveness of exposure. Bees react less strongly to a pulsed EF. In this case their level of excitation decreases with increased on-off time. The mechanismsof perception of the low-frequency EF is related to the physiological influence of the applied currents passing through the places of contact of the bees with each other or with the current-conducting surfaces. Thanks to the fact that the EF has a strong influence on bees, it can be used as an effective means for controlling their behavior.

[Text] Problems of the generation, perception and use of electric fields (EF's) by animals are receiving increasingly greater attention from specialists of different fields of knowledge. Interest in this area is caused by the fact that recent studies have shown the wide possibilities for using EF's as a means of regulating certain physiological processes and controlling the behavior of animals (Presman, 1968, 1974; Protasov, 1972, 1973; Kholodov, 1975, and others).

In terms of the problem under consideration, the question of the response of insects to EF's had been little studied, although even here some interesting information had been obtained. Thus, it had been established that insects generate (Gulyaev et al, 1970; Yes'kov, Sapozhnikov, 1974) and perceive (Altmann, 1959; Edvards, 1960; Husing et al, 1960; Yes'kov, 1967, 1969; Yes'kov and Sapozhnikov, 1975, 1976) EF's. Specific reactions of gregarious bees (Husing et al, 1960; Yes'kov, 1969, 1973; Yes'kov and Sapozhnikov) and wasps (Husing et al, 1961) to alternating EF's in relation to high intensities have been demonstrated.

The present work is devoted to study of the response of honeybees to low-frequency EF's. Our objective was to observe the action of EF's on bees in connection with the properties of their gregarious way of life. We also studied the mechanisms of perception and the possibility of using EF's for controlling the behavior of these insects.

The investigations were carried out on bee colonies, small groups of bees and separate individuals. The experimental insects were placed between two metal sheets, united with the high-voltage coil of a transformer. Its low-voltage coil was fed from an oscillator (GZ-3) across a power amplifier. In a number of experiments a semiconductor voltage converter was used as a high-voltage source. It made it possible to obtain an intensity of as high as 5 kV at a frequency of approximately 500 Hz.

The level of excitation of the bee colonies under the influence of the EF's was judged by the change in the sounds they made and by the increase in temperature and the concentration of carbon dioxide in the hive. The sounds were recorded with a microphone (MD-59) which was attached to the top of a plywood pyramid placed in the space above the hive. A special selective amplifier (Yes'kov et al, 1976) was used as a sound spectrum analyzer. It was used to record change in the intensity of the spectral components in the region of 440-460 Hz, a rise in which indicated increased activity of the bees (Yes'kov, 1970, 1972). In order to record the temperature we used an MKMT-16 microthermometer. It was located in the center of the space above the hive. The concentration of carbon dioxide was determined using OA-2209 and ATZ2CO$_2$ gas analyzers.

The action of the EF on groups of bees removed from the colony or on separate individuals was evaluated according to the quantity of oxygen required by them (MN-5124) and carbon dioxide given off (OA-2209 unit) and by the change in the integral integrity of the sounds. A condensor storage unit with a constant time of approximately 100 sec was used for integration.

Change in the bee's sensitivity to the action of the current in dependence on its frequency was studied in individuals which frequented a feeder with a 60-percent sugar solution. A GZ-3 generator was used as an electric stimulator. The rating of the current passing through the bee was recorded using an LV-9 millivoltmeter. The generator-voltmeter circuit was closed when the bee immersed its proboscis in the capillary containing the sugar solution. The amplitude of the current in this circuit was gradually increased until the bee stopped sucking the syrup (withdrew its proboscis).

Results and Discussion

The action of an EF on bee colonies stimulates excitation of adult individuals. The level of excitation depends on the intensity and the frequency-time structure of the field. The first indices of the activation of the bees under the influence of the field at a frequency of 100-1000 Hz was observed at an intensity of 5-14 V/cm[1], which can be recorded only by the acoustic method (fig 1) [figures not reproduced].

At relatively high intensities of the EF, excitation of the workers of the bee colony involving change in the microclimate of the hive was observed. A small change in the temperature (by 0.1-0.3 degrees) and the concentration of carbon dioxide (by 0.05-0.1 percent) in the hive is observed under the influence of a low-frequency EF at an intensity of 15-18 V/cm. Significant changes in the examined indices of the microclimate of the hive occur under the influence of fields with an intensity of tens of volts per centimeter. Here, the greater the intensity, the more the temperature and the concentration of carbon dioxide increase (fig 1).

The temperature and the gas composition of the bees' habitation change uniquely in dependence on the duration of the influence of the EF. For example, at one-minute operation of an EF at an intensity of 35 V/cm (frequency 500 Hz) the maximum increase in temperature is 1.2 degrees, and the concentration of carbon dioxide is 0.7 percent (fig 2A). Five-minute stimulation of the colony with the same field provokes a 2.7 degree increase in temperature and a 1.7 percent increase in the concentration of carbon dioxide (fig 2B). The action of this stimulus for one hour is expressed in a 3.6 degree increase in temperature and a 2 percentincrease in the concentration of carbon dioxide. The return of the temperature and the concentration of carbon dioxide to the initial level begins as early as the period of the influence on the colony of the EF, 18-23 and 8-10 min after it is switched on (fig 2B). This is connected with a sharp increase in the number of bees engaged in aerating the habitation. Increased aeration of the central zone of the nest--the place where the bees' brood is located--may also explain why it is in this part of the bee habitation that the temperature increases least of all under the influence on the colony of relatively high intensity EF's (Yes'kov, 1972).

It is important to note that despite the clear and pronounced tendency for the temperature and concentration of carbon dioxide in the hive to return to their initial levels as early as the period of prolonged action of the EF, the bees nonetheless adapt poorly to its influence. This is indicated by the high intensity level of sounds during the entire period of operation of the field. Their intensity returns to the initial level only some time after the EF has been shut off. The greatest stimulation of the workers of the colony under the action of the EF occurs during the first four to seven minutes after the EF is switched on. This fact is suggested by the maximum intensity of sounds during the indicated period (figs 1, 2).

Compared with a continuous EF operating for an equal interval of time, a pulsed EF does not stimulate an increase in the bees' activity. To the contrary, pulsed fields activate the bees more weakly. Thus, under the influence of a continuous EF operating for one minute (frequency 300 Hz, intensity 79 V/cm), the temperature over the nest of the experimental colonies increases on the average by 2.1±0.4 degrees and the concentration of oxygen decreases by 0.9±0.14 percent. This same stimulus fed in pulsed operation with an on-off time (the relation of the period of succession of the impulses to their duration) of 2 and an impulse duration of approximately 30 ms caused an increase in temperature by 1.2±0.2 degrees and a decrease in the concentration of oxygen by 0.5±0.1 percent. With an on-off time of 10 these indices of the microclimate of the hive changed by 0.4±0.1 degrees and 0.23±0.07 percent all told, respectively.

The level of excitation of the bees under the influence of EF's depends on their quantity. According to the data of analysis of the intensity of respiration, not one isolated individual reacted to the EF. The maximum intensity of the field to which the bees were exposed was 550 V/cm. The intensity of the respiration of two bees under the influence of the EF at the same intensity increased 1.5-fold (C_V=28 percent); of ten bees--1.8-fold (C_V=22 percent). With respect to the intensity of sounds, the first signs of excitation of 100 bees under the influence of an EF at a frequency of 300-600 Hz was observed at an intensity of 80-200 V/cm. This exceeds more than ten-fold the intensity necessary to excite a colony containing 15-20 workers.

The activating action of EF's on bees can be used as an effective means for controlling their behavior. In particular it is used for controlling swarming (Yes'kov, 1973), which involves the accumulation of passive bees in the hive. The influence of the EF on a colony in late-summer-early fall stimulates an increase in its brood. Thus, a colony subjected daily during the first half of September to 10-minute stimulation by an EF at an intensity of 100 V/cm (frequency of approximately 500 Hz) increased on the average by 3,200 workers by the end of the month (fig 3). This is nearly 30 times higher than the number of bees raised by the colonies of the control groups. (the experimental and control groups each contained 10 bee colonies.)

The above account indicates the strong physical action of low-frequency EF's on bees. Also noteworthy is the specificity of their response to this stimulation, which differs from their reaction to other stimuli, such as light, chemical substances and sound, which are perceived by specialized receptors.

The specific response of the bees to EF's can be explained by the properties of their perception of this stimulus. A hypothesis was recently proposed on this subject, according to which one of the mechanisms of perception of EF's involves the action of applied currents passing through the points of contact of the bees with eachoother or with other current-conducting surfaces (Yes'kov, Sapozhnikov, 1976). The results of the present work confirm this hypothesis. In fact, the operation of the frequency dependence of the change in the bees' activity under the influence of the EF (fig 4) can be explained by comparing the ratings of the applied currents at the corresponding frequencies and field intensities (fig 4A) with the change in the bees' sensitivity to currents of different frequencies. Thus, according to the reference data, the increase in the magnitude of the current applied to the bee at a field intensity of 500 V/cm is approximately 0.53 μA per kHz. The frequency dependence of the bees' sensitivity to the current[2] is clearly distinguished by pronounced irregularity. In the range of 0.05-0.2 Hz the sharpness of the drop in sensitivity is approximately 0.13 μA kHz; in the frequency range of 0.2 to 0.5 Hz it is 0.4 μA/kHz, and in the ranges 0.5-1.0 and 1.0-2.5 kHz it is 0.56 and 0.59, respectively. Owing to this the relatively small drop in the bees' sensitivity to the current in comparison with its increase in magnitude in the range of 0.05-0.5 kHz results in an increase in the level of stimulation under the influence of an EF of equal intensity. With further increases in frequency the increase in the rating of the applied current is compensated by a drop in the bees' sensitivity to it. In line with this the level of their activation decreases (fig 4a).

Further confirmation of the fact that the bees' perception of the EF is related to the applied currents is the diversity of forms of the frequency dependencies of comparable levels of stimulation (normalized in relation to the maximum) under the influence of fields at intensities of 150 and 500 V/cm (fig 4a). This is explained by the increase in the diversity of the currents applied in fields of different intensities with increase in their frequencies. In fact the greater the intensity of the EF, the more strongly the amplitude of the the applied current increases with increase in the frequency of the EF in relation to the threshold of sensitivity (fig 4b).

From the perspective of the stated concept of the perception of alternating EF's, it is possible to explain the increased excitation of the bees which occurs with increase in their number in the group undergoing stimulation by the field. Here the fact is that with increase in the number of individuals the number of tactile contacts between them increases. In this way the bees are subjected more frequently to stimulation by the current.

FOOTNOTES

1. In all cases the relation of the amplitude of the voltage applied to the sheets, to the space between them was taken as the intensity rating.

2. The method used inorder to determine the frequency dependency of the bees' sensitivity and the current gives one no idea of the absolute threshold of significance, since only a fairly strong stimulation can repell them from the food source. Therefore in the cases under examination only one aim was pursued--tc compare the ratings of currents of different frequencies which stimulate analogous reactions.

BIBLIOGRAPHY

1. Gulyaev, P. I.; Zabotin, V. I.; Shlippenbakh, N. Ya; Gorodiyenko, V. A. DOKL AN SSSR, Vol 191, 1967.

2. Yes'kov, Ye. K. VESTN N-I IN-TA PCHELOVODSTVA, Vol 17, Moscow, Moskovskiy Rabochiy, 1969.

3. Yes'kov, Ye. K. VESTN N-I IN-TA PCHELOVODSTVA, Vol 14, No 4, Moscow Moskovskiy Rabochiy, 1969.

4. Yes'kov, Ye. K. VESTN N-I IN-TA PCHELOVODSTVO, Vol 19, Moscow, Moskovskiy Rabochiy, 1970.

5. Yes'kov, Ye. K. ZOOL ZH, Vol 51, No 7, 1972.

6. Yes'kov, Ye. K. "Methods of Preventing Swarming of Bees", (Avt. Svid.) No 390797, in OTKRYTIYA IZOBRETENIYA, PROMYSHLENNYYE OBRAZTSY, TOVARNYYE ZNAKI, Vol 31, 1973.

7. Yes'kov, Ye. K.; Sapozhnikov, A. M. ZOOL ZH, Vol 53, No 5, 1974.

8. Yes'kov, Ye. K.; Sapozhnikov, A. M. DOKL VASKhNIL, Vol 4, 1975.

9. Yes'kov, Ye. K.; Sapozhnikov, A. M. BIOFIZIKA, Vol 21, No 6, 1976.

10. Yes'kov, Ye. K.; Sapozhnikov, A. M.; Toroptsev, A. I. PCHELOVODSTVO, No 4, 1976.

11. Presman, A. S. "Elektricheskiye polya i zhivaya priroda" [Electric Fields and Nature], Moscow, Nauka, 1968.

12. Presman, A. S. "Elektromagnitnaya signalizatsiya v zhivoy prirode" [Electromagnetic Signalling in Nature], Moscow, Sovetskaya Radio, 1974.

13. Protasov, A. P. "Bioelektricheskiye polya v zhizni ryb" [Bioelectric Fields in the Life of Fish], Moscow, Nauka, 1972.

14. Kholodov, Yu. A. "Reaktsiya nervnoy sistemy na elektromagnitnyye polya" [The Reaction of the Nervous System to Electromagnetic Fields], Moscow, Nauka, 1975.

15. Altmann, G. Z BIENENFORSCHUNG, Vol 4, No 10, 1959.

16. Edvards, D. K. CANAD J ZOOL, Vol 38, No 5, 1960.

17. Husing, J. O.; Struss, F.; Weide, W. NATURWISSENSCHAFTEN, Vol 47, No 1, 1960.

18. Husing, J. O.; Struss, F.; Weide, W. NATURWISSENSCHAFTEN, Vol 48, No 1, 1961.

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CSO: 184C

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EFFECT OF A PERMANENT MAGNETIC FIELD ON ELABORATION OF CONDITIONED REFLEXES IN MICE UNDER FOOTHILL AND ALPINE CONDITIONS

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/Article by T. Ryskanov and Z.M. Abdullina, Chair of Physics of the Kirghiz State Medical Institute, Frunze/

/Text/ During a study of the effect of a permanent magnetic field (PMP) on the conditioned reflex activity, it was shown that the PMP largely inhibits the appearance of conditioned reflexes. The development of tissue hypoxia is observed simultaneously (3, 8, 9/. Under pressure chamber and alpine conditions, it is observed that the effect of a moderate oxygen deficiency is accompanied by serious disturbances of the higher nervous activity /5/.

We did not find, in the literature, papers devoted to the study of the combined effect of a PMP and factors of an alpine environment on the conditioned reflex activity. At the same time, it is well known that adaptation to the slight effect of any factor increases the resistance of the organism to a more powerful effect /4, 6/.

In connection with this, we faced the problem of studying the combined effect of PMP and factors of an alpine environment on conditioned reflexes of avoidance.

Stimulation by an electric current and elaboration of conditioned reflexes of avoidance in white rats were performed in a T-shape labyrinth /7/. The labyrinth corridor was 50 cm long and 10 cm wide, the length of each branch was 20 m and the height of the labyrinth was 5 cm. Within 3-4 days the animals conducted 10 correct runs in the course of the experiment without stimulation by electric current or after 1-2 initial stimulations.

The magnetic field (MP) was created by a permanent magnet OMR-6 /expansion unknown/. Diameter of the magnet poles and the interpolar spaces were 80 mm.

We determined the field intensity of the MP with an IMI-1 /Izhevsk Mechanical Engineering Institute/ type gauge which operates on the basis of the Hall effect. We used a PMP intensity of 2000 ergs with a gradient of 200 ergs per cm, 200 ergs with a gradient of 25 ergs per cm and 20 ergs with a gradient of 1 erg per cm.

At the time of effect of the PMP, the experimental mice were placed in special chambers made from transparent plastic. Control animals were placed under the same conditions but were subjected to only the "imaginery" effect of the PMP. Experiments were performed on 92 mice in 6 series (3 in a foothill environment and 3 under an alpine environment).

In the first series of the experiment, under foothill conditions, 7 experimental mice and 7 control mice were subjected to a 2-hour exposure to a PMP of 2000 ergs intensity at one and the same time daily before the experiment. The use of this regime greatly increased the number of reinforcements necessary for emergence of the conditioned reflex of avoidance (Figure 1) and increased the time of a run in the "safe" branch of the labyrinth (from 2 ± 0.5 to 3.5 ± 0.3 sec; R 0.05). We must emphasize that all animals of the experimental group displayed a significant increase in the number of incorrect runs on the 2d-4th day. Then there was observed a repression of the conditioned reflex of avoidance and, afterward, the experimental mice required greater reinforcement than the control group to achieve the criterion of elaboration.

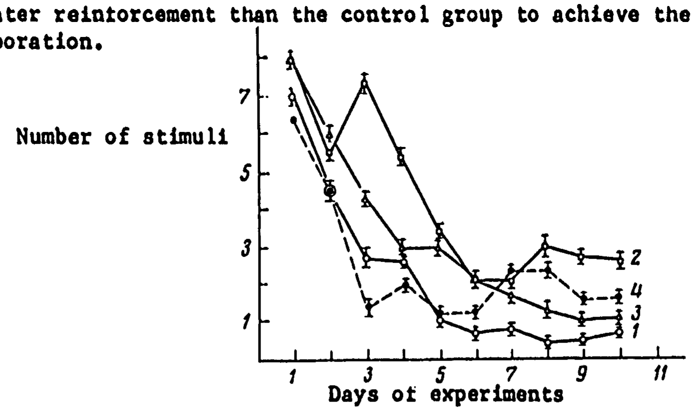

Figure 1. The effect of PMP of different intensity on the elaboration of a conditioned reflex of avoidance in mice under the foothill environment. 1 - control mice, 2 - PMP of 2000 ergs; 3 - 200 ergs, 4 - 20 ergs. Confidence intervals are shown in the brackets

In the 2d series of experiments (10 experimental mice and 10 control mice), the experimental animals also were subjected to 2-hour exposure to a PMP of 200 ergs intensity for 10 days before each experiment. Throughout the observations, the experimental animals required more reinforcements for achieving the criterion of elaboration of the conditioned reflex of avoidance in comparison with the control group (Figure 1).

In the 3d series of experiments (8 experimental mice and 8 control mice) we used a PMP of 20 ergs intensity. In the course of 5-6 days, the indicators of elaboration of a conditioned reflex of avoidance in the experimental animals did not differ from those in the control mice (Figure 1). After the 6th day of the experiments, the strength of the conditioned reflex diminished noticeably. These data justify the assumption that MP (Magnetic field) of this intensity produces an effect only after 5-6 days, that is, the organism of the animals begins to react to a given intensity only after summation of several exposures. Thus, the effectiveness of the effect of the PMP depends on the MP intensity and the duration of exposure.

Analogous experiments for the study of the conditioned reflex activity of mice were conducted under conditions of the combined effect of PMP of the indicated intensities and factors of the alpine environment (Tua-Ashu pass, 3200 m above sea level). Each series of experiments included 10 experimental animals and 10 control animals. Results of these series of experiments are shown in Figure 2. The use of a PMP of 20 ergs intensity did not affect the conditioned reflex of avoidance in the course of 5 days. From the 5th day to the end of the observations (up to 10 days), conditioned reflexes of avoidance were inhibited in comparison with the control. Evidently, PMP of this intensity has an effect only through 5-6 days during daily 2-hour exposure.

After use of a PMP of 200 ergs, the animals were trained best in comparison with control animals and with animals of the experimental group which underwent exposure to the 20 erg PMP. The mice were trained even better under the use of 2000 ergs field intensity.

We may conclude, on the basis of data obtained, that high intensities of PMP facilitate reduction of the reaction of the central nervous system of animals to alpine factors but a weak intensity PMP affects the adaptational capacities of the organism to a lesser degree.

Our data concerning the inhibiting effect of a PMP on elaboration of a conditioned reflex agrees with data of other researchers /2, 3, 8, 9/. Results of many studies /1, 3 and others/ indicate that amplification of the intensity of the PMP increases functional changes in the organism. The complex of stimulations associated with the stay of the organism under alpine conditions causes inhibition of elaboration and reinforcement of conditioned reflexes. The isolated effect of a PMP also causes similar reactions. However, the joint effect of a PMP and factors of an alpine environment leads to normalization of elaboration and reinforcement of conditioned reflexes.

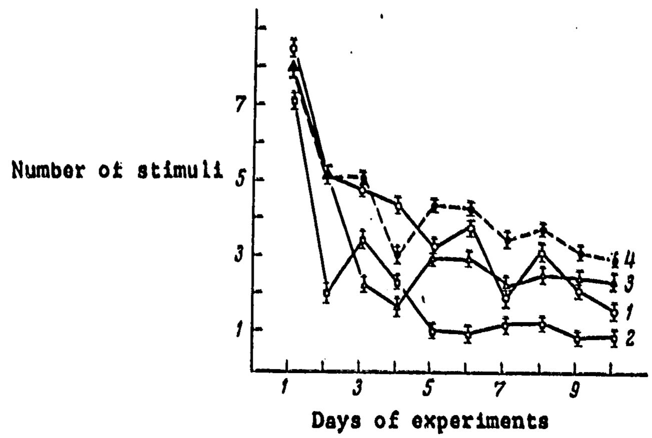

Figure 2. Effect of PMP of different intensity on elaboration of a conditioned reflex of avoidance in mice under alpine conditions. Symbols same as in Figure 1.

Consequently, the externally uniform reactions to a PMP and factors of the alpine environment revealed appear as antagonists during the joint effect of them.

BIBLIOGRAPHY

1. Abdullina, Z.M., Biological Effect of Magnetic Fields on the Animal Organism, Frunze, "Kyrgrystan," 1975.

2. Aminev, G.Ya., The Effect of a Permanent Magnetic Field on Some Inhibiting Processes. Candidate's Dissertation. Perm Medical Institute, 1966.

3. Asabayev, Ch., Materials on the Study of the Sensitivity of the Central Nervous System of Animals to Superhigh Frequency Electromagnetic Fields, Candidate's Dissertation, M., Institute of VND and NF of the AS USSR, 1971.

4. Barabashova, Z.I., KOSM. BIOL. I MED. (Space Biology and Medicine) No 4, p 6, 1969.

5. Meerson, F.Z. and Isabayeva, V.A. ZH. VYSSH. NERV. DEYST. (Journal of the Higher Nervous Activity) No. 2, V 21, 1971, p 470.

6. Ukolova, M.A., Kvakina, Ye.B. and Mar'yanovskaya, G.Ya., Materials of the Second All-Union Conference on the Study of the Effect of Magnetic Field on Biological Objects, M, p 237, 1969.

BIBLIOGRAPHY (continued)

7. Ryskanov, T. and Abdullina, Z.M. In: Magnetic Field in Medicine, Frunze, Central Committee of the Kirghiz Communist Party Publishing House, V 100, p 40, 1974.

8. Kholodov, Yu.A., The Effect of Electromagnetic and Magnetic Fields on the Central Nervous System. Moscow, NAUKA (Science) 1966.

9. Kuzman, E., Rulyhe, I, and Mesraros L., The Effects of Magnetic Fields on the Central Nervous System, Europ. Biophys. Congr., Vinne, V 5, p 253, 1971.

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CYCLES OF RESTORATION OF EVOKED POTENTIALS OF RAT BRAIN UNDER THE EFFECT OF A PERMANENT MAGNETIC FIELD

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/Article by N.P. Smirnova, Moscow/

/Text/ Some experiments have shown that the effect of a permanent magnetic field (PMP) leads to generalized changes of the bioelectrical activity of the brain /2, 4, 6-9, 13-15/. However, interpretation of the functional significance of changes revealed by different authors under different experimental conditions was not always the same. The nature of the changes under conditions of the general effect of a PMP of H 1000-4000 ergs observed by us (shifts of the frequency characteristics of background electrograms of different sections of the brain toward more rapid oscillations, results of some functional loadings and the appearance of evoked potentials complicated in form and increased in amplitude) justified the assumption that they reflect an increase of functional activity of nerve structures /2,6/. This assumption agrees with some data of the literature but requires experimental confirmation. One of the indicators of the functional state of the brain is the level of excitability of its structures.

In this study we determined the level of excitability of the cerebral cortex, the hypothalamus and the cerebellum cortex.

PROCEDURES. Experiments were performed on 20 white rats, anesthesized by nembutal (40 mg/kg, intraperitoneally). Evoked potentials (VP) were abducted from the sensory motor section of the cerebral cortex, the anterior-medial sections of the hypothalamus, the cortex of the anterior cerebellar vermis and were registered on a VC-7 oscillograph. We used silver electrodes for abduction from the cortical sections and Nichrome for abduction from the hypothalamus (diameter of the electrode tip -- 100-120 MKM). An indifferent electrode in the form of a non-magnetizing metal clamp was fastened to the skin incision. We stimulated the sciatic nerve from an MSE-40 stimulator by square paired pulses of 0.5 msec at a voltage twice the threshold voltage (2-6 v), which is usual for conditioning and testing stimuli. We conducted the paired stimulation program with gradual increase of the interval between stimuli within the limits of 20 up to 120 msec before, during and after the effect of the PMP. During processing of results, we considered the form and amplitude of the VP measured from peak to peak in the part of the potential with the highest amplitude. For each stimulus, we registered 6-10 responses, the amplitudes of which we averaged, at first in individual moments of each experiment and then for all rats. We determined localization of the electrodes in the hypothalamus by the method of photographing native sections of the brain.

We created the permanent magnetic field in an SP-15A electromagnet with plane parallel 400 x 300 mm terminals and a distance of 100 mm between them. The voltage pulsation equals 1.8 percent. The magnetic field was practically homogeneous in the central part of the 300 x 200 mm interpolar space, in which we placed the rats, fixed on a stand. Thus, the animals were subjected to the total effect of the PMP during vertical passage of the lines of force.

RESULTS OF THE STUDY AND DISCUSSION OF THEM

We pointed out earlier /2/ that, during a stay in a PMP of sufficiently high intensity (500-1000 ergs and higher), there occurred in rats, instead of the 2-phase VP usually registered in different structures of the brain upon stimulation of the somatic nerve, high-amplitude multi-component potentials.

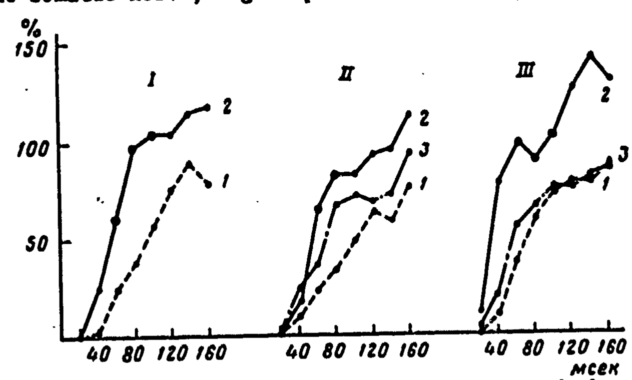

Figure 1. The effect of PMP on the restoration of the evoked potential of the brain on the testing stimulation (mean value) of the nerve. Along the ordinate axis -- the amplitude of the testing VP, % for the amplitude of the conditioning VP; along the axis of the abscissa -- interstimuli intervals, μs. 1 - sensomotor cortex; 11 - hypothalamus; 111 - cerebellum cortex, 1 - before effect; 2 - in PMP H 500-1000 ergs; 3 - after effect of PMP H 4000 ergs. Thick points differing reliably from the control.

Therefore, for registration of evoked activity under conditions of paired stimulation of the sciatic nerve at the time of effect of the PMP, in each experiment, an intensity (500-1000 ergs) was selected under which pronounced changes of the shape of the VP did not arise in a given rat. Then, after paired stimulation, we increased the field intensity up to 4000 ergs and, within 15-30 minutes, we switched off the electromagnet and repeated the stimulation program anew. Thus, rats were subjected to a single, 2-stage effect of PMP H 500 or 1000 and 4000 ergs during a total exposure of 1 - 1½ hours.

As is well known, the cycle of restoration of VP (reactions to testing stimuli) involves three basic periods -- areactivity, subnormality and supernormality. The duration of the individual periods and of the entire cycle varies widely in dependence upon the magnitude of the conditioning and testing stimuli, the functional state of the animal and other conditions of the experiment /3, 5, 10-12, 17/. Under the stimulation program selected up to the effect of the PMP in rats under deep nembutal narcosis in all three structures of the brain, the restoration of response to the testing stimulation of the nerve began at intervals between stimuli of 40-60 msec; according to mean data the duration of the absolute refractory phase lasted 50 - 64.6 msec, later the amplitude of the response gradually increased and at the longest interval studied (160 msec) reached 76-89 percent of the amplitude of the conditioning potential (Table, Figure 1). These data are comparable to results obtained during study of cycles of restoration of VP of the cerebellum vermis to stimulation of the sciatic nerve in adult immobilized rats. The first signs of restoration of the testing VP appeared at an interval of 33 msec and complete restoration appeared at an interval of 145 msec. Further increase of the inter-stimulus interval up to 1 second did not change the amplitude of the testing potential.

Duration of the Absolute Refractory Phase, msec

Условия опыта	Сенсомоторная кора	Гипоталамус	Кора мозжечка
До воздействия	64,6±11,7	63,1±11,7	50,0±4,8
В ПМП	40,0±4,5 $p = 0,05$*	51,4±19,7	17,1±2,8 $p < 0,001$
После воздействия	Не исследовали	28,0±3,8 $p < 0,02$	33,3±4,5 $p < 0,01$
Число крыс	12	8	10

* p - Comparison with control

Key: 1. Conditions of experiment
2. Sensomotor cortex
3. Hypothalamus
4. Cerebellum cortox
5. Before effect
6. In PMP,
7. After effect
8. Not studied
9. Number of rats

The effect of the PMP caused significant changes of cycles of restoration of the VP of the rat brain. During a stay of the animal in a "subthreshold" field intensity there occurred, in comparison with initial data, more rapid and more complete restoration of response to the testing stimulation of the nerve and during this, the duration of the absolute refractory phase (period of areactivity) and the period of subnormality were reduced accordingly. Less pronounced changes of cycles appeared immediately after cessation of the effect of the H 4000 erg field. In the time of effect of the PMP, it was possible to observe, in addition to the shortening of periods of areactivity and subnormality, in some experiments in the process of restoration, the phenomenon of supernormality -- alleviation of the testing response.

In the experiment illustrated by Figure 2, the effect of the H 1000 erg PMP caused an increase of the amplitude of the conditioning and the testing responses without a change of the form of the potential in both structures, the sensomotor region of the cerebral cortex and the anterior hypothalamic nucleus. Complete restoration of cortical response to the testing stimulation occurred during an inter-stimulus interval of 60 msec. In the hypothalamus, at this moment, the amplitude of the testing potential before the effect constituted approximately one-half of the amplitude of the conditioning response and in the PMP exceeded it. Further increase of the intervals is accompanied by alleviation of the second response in both structures and is more pronounced at the time of effect of the PMP.

Changes of cycles of restoration of VP were most significant in the cerebellum cortex. This is seen clearly by the mean data. In the cerebellum cortex, in comparison with the cerebral cortex and the hypothalamus, there was a more abrupt decrease of duration of the absolute refractory phase of the cycle of restoration (Table), and the VP amplitude to the second stimulation of the nerve at 120-160 msec intervals between stimuli reached 125-142 percent of the amplitude of the conditioned response. In the cerebral cortex and the hypothalamus, supernormality was characterized at 117 and 114 percent (Figure 1).

Changes of cycles of restoration of VP observed and, notably, the contraction of the absolute refractory phase, the shortening of the period of supernormality, phenomena of alleviation indicate an increase of excitability of the cerebral cortex, the hypothalamus and the cerebellum cortex under the effect of a permanent magnetic field. The shortening of cycles of restoration of VP is consistent with the quickening of spontaneous bioelectrical activity under the effect of a PMP /6/. As is well known, the duration of cycles of excitability increases during predominance in the background rhythm of slow oscillations and is shortened if rapid rhythms predominate in the background /5/.

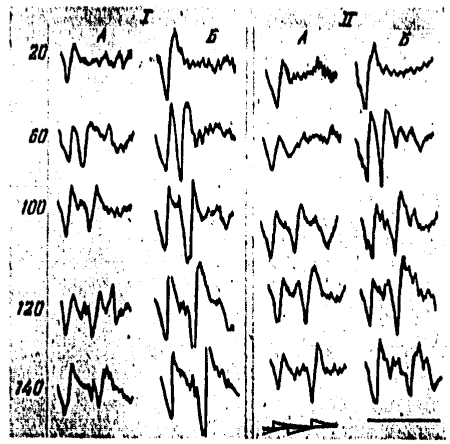

Figure 2. The effect of a PMP H 1000 ergs on restoration of the VP of the cerebral cortex (1) and the hypothalamus (II) to a testing stimulation of the sciatic nerve. A -- before the effect; B - in the PMP. Figures represent the intervals between stimuli, msec. Calibration of amplification 50 mkv, Time marking 10 msec.

There is also great interest in the possibility of amplification or even provocation (appearance in the VP structure of supplemental oscillations) of the magnetic effect as a result of paired stimulation of the sciatic nerve. This effect was noted in 6 rats in all 3 sections of the brain and was developed at an interval between stimuli beginning from 40 - 60 msec, which corresponds to the frequency of stimulation of 25-16 pulses per second.

In the experiment, illustrated by Figure 3, (1, A and B), before the effect of the PMP, restoration of the testing response of the cerebellum cortex was obvious at an interval of 60 msec and reached complete expressiveness upon further increase of inter-stimuli intervals. Placement of this rat in the PMP of H 500 ergs led to changes of evoked activity and in response to paired stimulation of the nerve at a 20 msec interval, a conditioned response and supplemental single-phase oscillation were registered. Upon increase of the inter-stimulus interval up to 60 msec and more, the effect of the PMP was amplified: the form of the potentials was complicated due to the imposition on the conditioning and testing responses of supplemental high-amplitude oscillations. In another experiment (Figure 3, II, B) in a PMP of the same intensity, no changes in the magnitude and form of VP of the cerebral cortex were observed in the beginning. However, beginning with an inter-stimulus interval of 100 msec, paired stimulation of the nerve led to provocation of the magnetic effect -- to the manifestation following the testing stimulus of a 3-phase oscillation, attaining, at an interval between stimuli of 140 msec (frequency of 7 pulses per second), a maximal amplitude (from peak to peak) of 380 mkv and the amplitude of the conditioning response equalled 100 mkv.

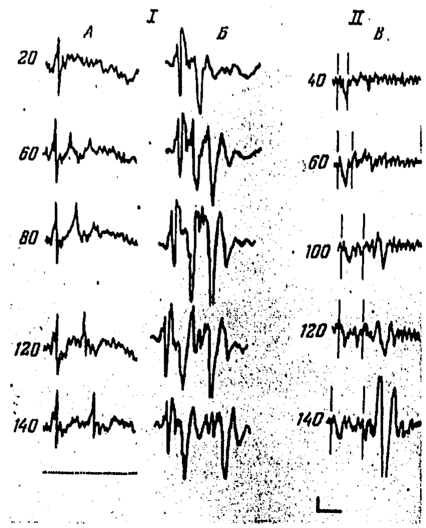

Figure 3. Amplification (1) and appearance (II) of the effect of a PMP on the VP of the cerebellum cortex under conditions of stimulation of the sciatic nerve by paired current pulses. A - before the effect; B and C - during the effect. The figures represent intervals between stimuli, msec. Calibration of amplification of 50 mkv, time marking 10 msec.

In all probability, the appearance or amplification of the magnetic effect facilitates the state of supernormality, being created in the nerve elements of the brain at the time of effect of the PMP. The phase of supernormality may be noted in individual animals, beginning with inter-stimulus intervals of 40-60 msec with a maximum at intervals of 120-160 msec, precisely the same as the pehnomenon of amplification or provocation of the magnetic effect (Figure 3) is being developed under conditions of paired stimulation of the nerve at the same frequency. It was shown that alleviation of responses of the visual cortex to rhythmic light stimulation was most pronounced at a frequency of flashes (2 - 2.5 pulses per second) equal to the cycle of excitability (400-500 msec). This may be explained by the summation of the trace of supernormality from the first stimulus with excitation from the latter /5/.

A study of the mechanics of reactions of reconstruction of the EEG /Electroencephalogram/ of the occipital region also revealed an increase of the after-effect from stimulus to stimulus during an interval between them of 200 msec, that is, a frequency of light flashes, near to the natural frequency of the after-effect discharge /1/.

Thus, in a permanent magnetic field of "subthreshold" intensity during paired stimulation of the nerve, there arise conditions not only for alleviating the testing response but also for exaltation of supplementary oscillations in the VP structure -- "a magnetic effect" which, evidently, may occur to a large extent due to an increase of excitability of the central nervous structures and also due to the number of nerve elements involved in the evoked reaction.

The state of high excitability of the cerebral cortex, the hypothalamus and the cerebellum cortex is revealed by evaluating the cycles of restoration in the field (H 500-1000 ergs) in the absence of changes of evoked activity and also immediately after more intense effect (H 4000 ergs), during which the magnitude of the form of somatosensory VP undergo significant changes.

The data obtained represent significant shifts for functional evaluation of bioelectrical processes which take place under the effect of a PMP and confirm the assumption concerning amplification of the exciting process in the animal brain, subjected to the effect of a PMP.

Conclusions

1. The effect of a permanent magnetic field leads to an increase of the level of excitability of the cerebral cortex, the hypothalamus and the cerebellum cortex which is indicated by the contraction of the periods of areactivity and subnormality in the cycle of restoration of evoked potentials of these sections of the brain.
2. Under conditions of the effect of a permanent magnetic field of "subthreshold" intensity, paired stimulation of the sciatic nerve at definite inter-stimulus intervals may cause amplification or provocation of the "magnetic effect" -- the appearance in the structure of evoked potentials of supplemental oscillations.

BIBLIOGRAPHY

1. Gusel'nikov, V.I. and Supin A.Ya., Rhythmic Acitivity of the Brain. Moscow, Moscow State University Publishing House, 1968.

2. Klimovskaya, L.D. and Smirnova, N.P., BYUL. EXSPERIM. BIOL. I MED. (Bulletin of Experimental Biology and Medicine) V 82, No 8, p 906, 1976.

BIBLIOGRAPHY (continued)

3. Kondrat'yeva, I.H. In "Contemporary Problems of Electrophysiology of the Central Nervous System." Moscow, NAUKA (Science) p 148, 1967.

4. Luk'yanova, S.N., ZH.VYSSH. NERVN. DEYAT (Journal of the Higher Nervous Activity) V 17, No 4, p 722, 1967.

5. Skrebitskiy, V.G., FIZIOL. ZH. SSSR (Physiology Journal of the USSR) V 46, No 12, p 1429, 1960.

6. Smirnova, N.P. and Klimovskaya, L.D. ZH. VYSSH. NERV. DEYAT, V 26, No 2, p 403, 1976.

7. Kholodov, Yu.A. The Effect of Electromagnetic and Magnetic Fields on the Central Nervous System, Moscow, NAUKA, 1966.

8. Kholodov, Yu.A. Reactions of the Nervous System to Electromagnetic Fields, Moscow, NAUKA, 1975.

9. Chizhenkova, R.A., BYUL. EKSPERIM. BIOL. I MED (Bulletin of Experimental Biology and Medicine) V 61, p 11, 1966.

10. Bishop G.H. Amer. J. Physiol., V 103, No 2, p 213, 1933.

11. Chang, H.T. J. Neurophysiol., V 14, No 1, p 95, 1951.

12. Evarts, E.V., Fleming, T.C. and Huttenlocker, P.R., Amer. J. Physiol., V 199, No 2, p 373, 1960.

13. Gualtierotti, T. In: Proc. 12th Internat. Astronaut. Congress, V 2, p 586, 1963.

14. Knepton, J.C. and Beisher, D.E., Aerospace Med., V 37, No 3, p 287, 1966.

15. Kuzmann E., Pelyhe, I. and Meszaros, I., Europ Biophys. Congr., Vienna, V 5, p 273, 1971.

16. Mares P. Acta biol. et med. ger., V 35, No 1, p 55, 1976.

17. Morin, M.G., Gastaut, H., Naquet, R. and Roger, M.A., J. Physiol. (France) V 43, No 4, p 820, 1951.

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